

Transformation

Requirements:

- TransGen® Trans5α Chemically Competent Cell
- LB broth
- iGEM DNA Distribution Kit Plates, Plasmid DNA or DNA ligation mix
- LB agar plates containing 15-100µg/mL antibiotic of choice
- Nuclease-free 1.5mL microcentrifuge tubes
- Nuclease-free 0.2mL PCR tubes
- Water bath of 42°C
- Shaking incubator of 37°C

Before Starting (if you need plasmids from iGEM DNA Distribution Kit Plates):

1. Punch a hole with a pipette tip through the foil cover into the corresponding well of the desired BioBrick part.
2. Add 10µL sterile deionized water, pipette up and down several times.
3. Transfer liquid from Step 2 into a 0.2 mL PCR tube.
4. Repeat Steps 2-3 twice.
5. Store liquid (BioBrick plasmid) at -20°C

Protocol:

1. Add 5-10µL plasmid or ligation system into 50µL fresh competent cells, which is contained in 1.5mL centrifuge tube. Then mix gently.
2. Incubate the tube on ice for 30 minutes
3. Heat shock in the water bath at 42°C for 45 seconds.
4. Incubate on ice for 2 min.
5. Add 450µL fresh LB broth into the tube.
6. Incubate for 1 hour under the condition of 37°C, 200rpm using a shaking incubator.
7. Spread 250µL liquid from Step6 on a LB agar plate, which contains appropriate antibiotics.
8. Incubate overnight at 37°C (about 12 hours, no more than 16 hours).